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Author Affiliation:

¹Department of Fisheries, Faculty of Agriculture, University of Port Harcourt, Choba, Rivers State, Nigeria

²Department of Animal and Environmental Biology, University of Port Harcourt, Choba, Rivers State, Nigeria

Corresponding Author

Department of Fisheries, Faculty of Agriculture, University of Port Harcourt, Choba, Rivers State, Nigeria
Email: henry.dienye@uniport.edu.ng

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Diversity of harmful diatom species in the upper reaches of Bonny Estuary, Niger Delta, Nigeria

Dienye HE^{1*}, Sikoki FD², Mmeribe VO¹

ABSTRACT

This research was carried out to assess the abundance and distribution of harmful diatoms in the upper reaches of the Bonny Estuary between December 2021 and November 2022. Three sampling stations were established using the ArcGIS tool. Microalgae species were sampled with 20µm mesh plankton net. Some physicochemical parameters were determined in situ, while nutrients were analyzed in the laboratory using the APHA 4500 Method. A total of 8 genera and 18 species, with four genera of centric diatoms (8 species) and four genera of pennate diatoms (10 species) were recorded. The least mean density value (318.33 CellsL⁻¹), was recorded for *Thalassiosira eccentrica* species, while *Cyclotella meneghiniana* recorded the highest density value (1146.67 CellsL⁻¹), for centric harmful diatoms. *Nitzchia vermicularis* recorded the least density value (602.34 CellsL⁻¹) and *Navicula amphibola* recorded the highest density value (2022.34 CellsL⁻¹) for pennate harmful diatoms across the sampled stations. The percentage composition recorded 64% for pennate diatoms and 36% for centric diatoms. Salinity and temperature showed a strong positive correlation with pennate diatoms and pH, nitrate and phosphate also showed a strong positive correlation, with harmful centric diatoms. Diversity indices revealed Shannon's index ranged between 2.77-2.86 and species evenness ranged between 0.89-0.91, while Margalef range value 2.59-2.60 were considered moderately stable. All species were evenly spread across all stations. Further research is necessary to develop effective management strategies that can mitigate the possible formation of blooms and their harmful effects in the upper reaches of the Bonny Estuary.

Keywords: Diatoms, distribution, abundance, Estuary, harmful species.

1. INTRODUCTION

The essential component of living organisms in bays, estuaries, creeks, deep oceans and sediments is phytoplankton. These microscopic creatures' organic compounds determine the essential primary productivity of aquatic habitats. Diatoms are the most important primary producers in both freshwater and marine habitats (Yool and Tyrrell, 2003). Diatoms are one of the most important types of marine phytoplankton and are distinguished by their siliceous cell wall

(frustule) (Mann, 1999). They can account for up to 45% of the total primary output in the ocean or 20–25% globally (Werner, 1977).

They form the base of the food web in many marine ecosystems; they are significant players in the biogeochemical cycling of C, N, P, Si, and biologically required trace metals (Sarthou et al., 2005). Their success implies that they have highly efficient and adaptable survival mechanisms and growth strategies. Diatoms are often utilized in water quality research because they are sensitive to various environmental factors, such as water acidification, eutrophication and climate change. They have also been employed as helpful markers in historical water quality studies (Schlüter et al., 2012). Diatoms are algae with unique, translucent cell walls comprised of hydrated silica that live in freshwater, saltwater and moist vegetation on land (Nabors, 2004).

There are two primary kinds of diatoms: Central diatoms (Centrales), which have radial symmetry, such as Cyclotella and pennate diatoms (Pennales), which have bilateral symmetry e.g., *Synedra* (Kadiri and Opute, 2013). Diatoms are responsible for about 40% of the primary production in marine ecosystems and account for up to 20% of global carbon fixation (Geider et al., 2001). They are the most abundant unicellular algae in the ocean and freshwater settings and they provide vital food and oxygen to heterotrophs in aquatic ecosystems. Diatom cell walls are made up of two halves of silica (Dolphine, 2008).

Some diatom species' success may also be explained by their ability to build long chains that are difficult or impossible for some grazers to consume. Their frustules also provide automated protection against some grazers because they must be disrupted with great power (Hamm et al., 2003). In addition to this automated defense, some diatoms create unsaturated aldehydes, which significantly limit zooplankton and other invertebrate reproductive success and thus, population expansion. Diatoms, unlike other smaller phytoplankton, require nutrient-rich conditions for growth and turbulence to keep them suspended. As a result, they are regularly found near coastal areas, where their effects on humans and marine food webs are more frequently documented (Justic et al., 1995).

Because of increased nutrient enrichment caused by urbanization, tourism, industrial wastes, desalination plants, agricultural operations and ballast water, coastal ecosystems are becoming more vulnerable to harmful algal blooms (HABs), particularly in confined coastal embayments (Anderson et al., 2002; Sellner et al., 2003). Harmful microalgal diatoms play critical ecological roles in virtually all aquatic environments (Middelburg et al., 2000). Considering their importance, high abundance and enormous diversity, it is surprising how little attention their effect on ecosystem productivity has received. There is no recorded information on harmful diatoms in the upper reaches of the Bonny Estuary.

2. MATERIALS AND METHODS

Study area

Description of study area

The Bonny Estuary is among the numerous low land coastline of the Niger Delta Complex. It is located between 4° 25" and 4° 50" N latitude and 7° 0" and 7°15" E longitude in Rivers State, Nigeria. It is short, extending 180km from its mouth. It is mainly brackish and consists of a main river channel and creek. The Bonny Estuary channel has the highest tidal flow among river systems which is mainly influenced by tidal movements (Dangana, 1985). The three geo-reference stations were set up along the estuary course using the ACRC GIS tool. The stations chosen include: Station 1 (Abuloma jetty), Station 2 (Kalio Ama) and Station 3 (Ogoloma).

Sample Collection, Preparation and Analyses

Surface water hauls were obtained in triplicate at each sample location by filtering 50 litres of surface water through a 20 µm phytoplankton net. Keep in plastic bottles for examination and identification. According to the IOC handbook, samples were preserved promptly with 2% formaldehyde. Temperature, salinity, TDS, pH and DO were measured in situ at the sampling site using a horiba water checker (Model: Extech D0700). Nutrient samples (PO₄ and NO₃) were evaluated in the laboratory using the APHA 4500 Method.

Enumeration of Harmful Diatom Species

Diatom microalgae were enumerated with Lackey Drop Micro-transect Counting Method (APHA, 1998). The sample was mixed well before sub-sampling a drip of 0.05 ml onto a glass slide in triplicate with cover slip. The volume processed was known and the numbers of observed Microalgae were known in a given volume, their abundance was counted at Low power objective microscope. Microphotographs of Harmful algae were taken by employing a camera that was fixed to the microscope. Reference materials: Hallegraaff, (1995) and Tomas, (1997) were used to identify the harmful diatom species.

Individuals per ml were calculated;

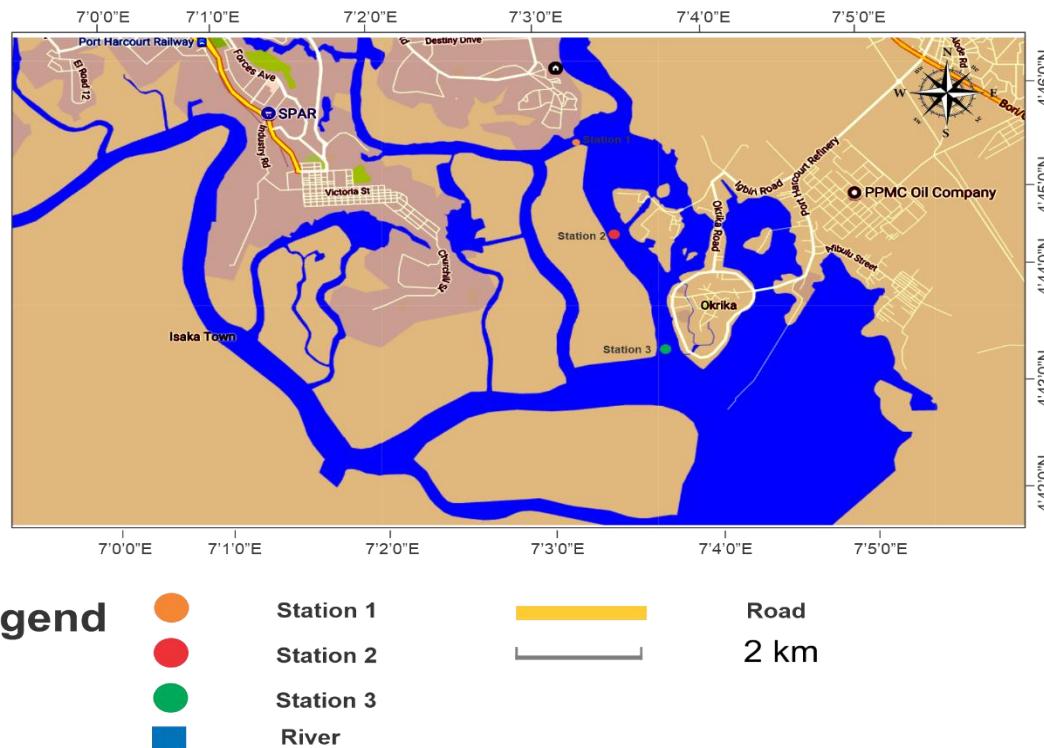


Figure 1 Map of the study area

$$\text{Number (No.) Individuals/ml} = \frac{C \times TA}{A \times S \times V}$$

Where, C = number of organisms counted; TA = area of the cover slip, mm²; A = area of one strip, mm²; S = number of strips counted and V = volume of sample under the cover slip, ml

Data Analysis

Analysis of Variance

The physico chemistry and nutrients were analysed using a one-way analysis of variance of SPSS v. 20. Tukey HSD was used to separate the mean differences at a 95% confidence interval (i.e., p<0.05).

Diversity indices

The diversity of harmful diatoms in the estuary was calculated using the PRIMER software version 6.1.6 (Clarke and Gorley, 2006) to enumerate:

Species diversity

Species abundance

Species distribution

Principal component analysis

Principal component analysis (PCA) was employed to analyze the relationship between harmful diatom species and significant environmental factors using PAST software (Leps and Similauer, 2003).

3. RESULTS

Table 1 shows the mean value of the environmental parameters of the upper reaches of the Bonny Estuary. There is no significant difference in all the physicochemical parameters across stations, nitrate shows a significant difference across stations (p>0.05) while there was no significant difference in phosphate across stations (p<0.05)

Table 1 Environmental parameters of the Upper reaches of Bonny Estuary across stations

Parameter	Station		
	1	2	3
Ph	7.99±0.51 ^a	7.93±0.46 ^a	7.77±0.45 ^a
Temp.(°C)	28.60±1.47 ^a	28.42±1.57 ^a	28.53±1.69 ^a
DO (mgL ⁻¹)	5.13±0.64 ^a	5.12±0.66 ^a	5.03±0.61 ^a
BOD (mgL ⁻¹)	1.95±0.48 ^a	1.68±0.59 ^a	1.63±0.48 ^a
TDS (mgL ⁻¹)	28.55±2.95 ^a	29.97±3.69 ^a	31.89±4.03 ^a
Salinity (ppt)	3.12±1.37 ^a	2.91±1.31 ^a	2.73±1.38 ^a
Nitrate (mgL ⁻¹)	1.92±1.01 ^b	5.59±1.48 ^a	4.18±1.81 ^{ab}
Phosphate (mgL ⁻¹)	0.19±0.21 ^a	0.59±0.19 ^a	1.14±0.76 ^a

Superscripts of the same alphabet are not significantly different ($p > 0.05$).

Superscripts of different alphabets are significantly different ($p < 0.05$).

Table 2 showed the harmful diatoms in the upper reaches of the Bonny Estuary. One class of significant groups of harmful diatoms was recovered in the samples: Bacillariophyceae. A total of 8 genera and 18 species with 4 genera of centric diatoms (8 species) and 4 genera of pennate diatoms (10 species). Families (*Stephanodiscaceae*, *Coscinodiscaceae*, *Triceratiaceae* and *Thalassionematacea*) of centric diatoms recorded two species each, while for pennate diatoms, families of *Bacillariaceae* recorded five species; *Naviculaceae* recorded three species while *Pinnulariaceae* and *Pleurosigmataceae* recorded one species each.

Table 2 Harmful Diatom in the upper reaches in Bonny Estuary

Class	Family	Species
Centric Harmful Diatoms		
Bacillariophyceae	<i>Stephanodiscaceae</i>	<i>Cyclotella Antigua</i> (W. Smith, 1853)
		<i>Cyclotella meneghiniana</i> (Kützing 1844)
Bacillariophyceae	<i>Coscinodiscaceae</i>	<i>Coscinodiscus granni</i> (L.F Gough,1905)
		<i>Coscinodiscus radiatus</i> (Ehrenberg,1840)
Bacillariophyceae	<i>Triceratiaceae</i>	<i>Odontella aurita</i> (C. Agardh 1832)
		<i>Odontella mobilis</i> (Grunow,1884)
	<i>Thalassionematacea</i>	<i>Thalassiosira eccentrica</i> (Cleve 1904)
		<i>Melosira moniliformis</i>
Pennate Harmful Diatoms		
Bacillariophyceae	<i>Bacillariaceae</i>	<i>Bacillaria paxillifera</i> (Muller&Hendy,1951)
		<i>Nitzchia sigma</i> (W. Smith, 1853)
		<i>Nitzchia vermicularis</i> (Hantzsch,1860)
		<i>Amphiprora sp</i>
		<i>Licmophora sp</i>
	<i>Naviculaceae</i>	<i>Navicula amphibola</i> (Cleve,1891)
		<i>Navicula dicephala</i> (Ehrenberg 1838).
		<i>Gyrosigma acuminatum</i> (Rabenhorst 1853)
	<i>Pinnulariaceae</i>	<i>Pinnularia undulata</i> (Sensu Cleve,1891)
Bacillariophyceae	<i>Pleurosigmataceae</i>	<i>Pleurosigma elongatum</i> (W. Smith 1852)

Table 3 shows the mean density per ml of harmful diatom species. In station 1, *C. granni* recorded the highest mean density value of 318.00CellsL⁻¹ and *C. radiatus* recorded the least value of 147.17CellsL⁻¹ for centric species while *N. amphibola* recorded 806.67CellsL⁻¹ as the highest and 295.67CellsL⁻¹ for *N. vermicularis* as the least for pennate species. In station 2, *O. aurita* recorded the highest density value of 602.00CellsL⁻¹ and *C. antigua* recorded the least density value of 136.00CellsL⁻¹ centric species while *Navicula cryptocephala* recorded 693.00CellsL⁻¹ as the highest and 306.67 CellsL⁻¹ for *P. elongatum* as he least density value for pennate species.

Table 3 Mean Density per Litre of Harmful diatoms of the upper reaches of Bonny Estuary

Species	Station		
	1	2	3
Centric			
<i>Odontella aurita</i>	0.00±0.00 ^a	602.00±19.93 ^a	442.33±1.16 ^b
<i>Odontella mobilensis</i>	0.00±0.00 ^a	431.67±103.87 ^a	591.33±85.17 ^a
<i>Coscinodiscus granii</i>	318.00±71.34 ^a	261.67±316.89 ^a	249.67±256.24 ^a
<i>Coscinodiscus radiatus</i>	147.67±129.07 ^a	0.00±0.00 ^a	250.33±98.15 ^a
<i>Cyclotella antiqua</i>	306.67±90.15 ^{ab}	136.00±122.59 ^b	397.67±51.94 ^a
<i>Cyclotella meneghiniana</i>	227.00±336.01 ^a	510.67±207.14 ^a	409.00±117.78 ^a
<i>Melosira moniliformis</i>	218.00±129.29 ^a	340.00±34.00 ^a	181.33±51.94 ^a
<i>Thalassiosira eccentrica</i>	159.33±153.84 ^a	159.00±137.69 ^a	0.00±0.00 ^a
Pennate			
<i>Navicula amphibola</i>	806.67±104.53 ^a	658.67±239.28 ^a	557.00±193.89 ^a
<i>Navicula cryptocephala</i>	840.33±104.31 ^a	693.00±221.71 ^a	0.00±0.00 ^a
<i>Gyrosigma acuminatum</i>	488.33±128.72 ^a	601.67±141.55 ^a	511.00±269.87 ^a
<i>Bacillaria paxillifera</i>	568.00±153.79 ^a	374.67±328.45 ^a	363.33±489.52 ^a
<i>Pinnularia undulata</i>	375.00±155.80 ^a	0.00±0.00 ^a	397.67±51.94 ^a
<i>Pleurosigma elongatum</i>	431.67±157.04 ^a	306.67±266.13 ^a	308.00±89.41 ^a
<i>Nitzchia sigma</i>	398.00±119.88 ^a	0.00±0.00 ^a	522.67±258.02 ^a
<i>Nitzchia vermicularis</i>	295.67±39.26 ^a	0.00±0.00 ^a	306.67±156.35 ^a
<i>Licmophora sp</i>	363.33±175.03 ^a	386.33±128.72 ^a	0.00±0.00 ^a
<i>Amphiprora sp</i>	0.00±0.00	431.67±387.63	511.00±171.00

In station 3, *O. mobilensis* recorded the highest value of 591.33CellsL⁻¹ and 181.33CellsL⁻¹ for *M. moniliformis* recorded the least value for centric species while *N. amphibola* recorded a value of 557.00 CellsL⁻¹ as the highest and 308.00 CellsL⁻¹ for *P. elongatum* as the least density value for pennate species. Figure 2 shows the percentage composition of harmful diatoms, pennate diatoms recorded 64%, while centric diatoms recorded 36%, respectively.

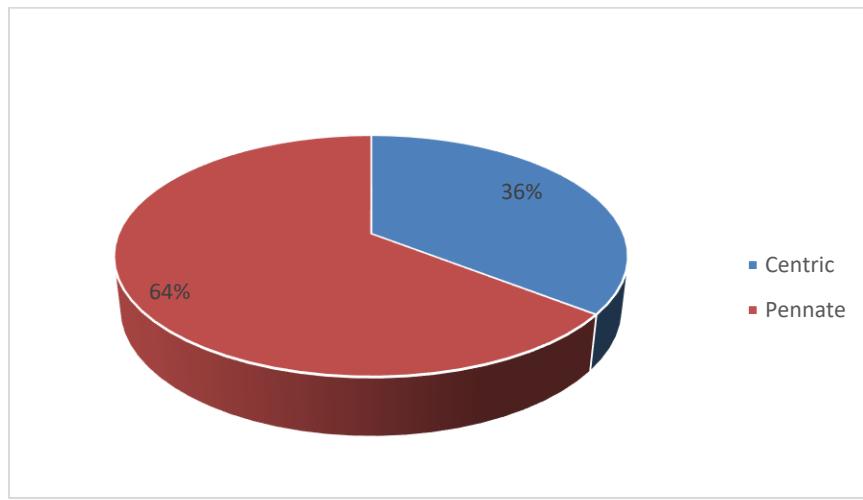
**Figure 2** Relative abundance /Percentage composition of centric and pennate harmful diatoms in Upper reaches of Bonny Estuary

Figure 3 showed the interaction between the environmental parameters centric and pennate harmful diatoms. Salinity and temperature showed a strong positive correlation with pennate diatoms while BOD and DO showed a weak positive correlation. pH, nitrate and phosphate showed strong correlation while TDS showed a negative correlation for harmful centric diatoms.

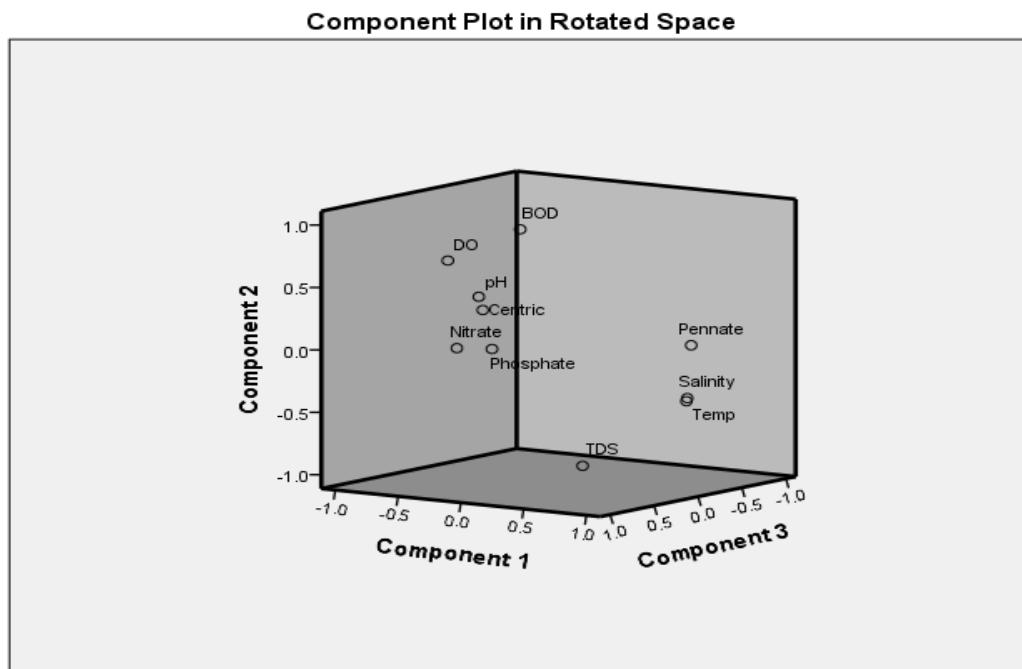


Figure 3 PCA Analysis of centric and pennate harmful diatoms with environmental parameters of the upper reaches of the Bonny Estuary

Figure 4 showed the interaction between the environmental parameters and harmful centric diatom species. *C. radiatus* showed a strong correlation with pH and slightly correlated with Dissolved oxygen, while *C. granni* showed a strong correlation with phosphate and nitrate. *C. menegiana* showed a very strong negative correlation with BOD while *C. antiqua* showed a strong correlation with TDS. *T. eccentrica* showed a strong correlation with salinity and *O. mobilensis* showed a strong correlation with phosphate. *M. moniliformis* and *O. aurita* showed strong negative correlation with the environmental parameters.

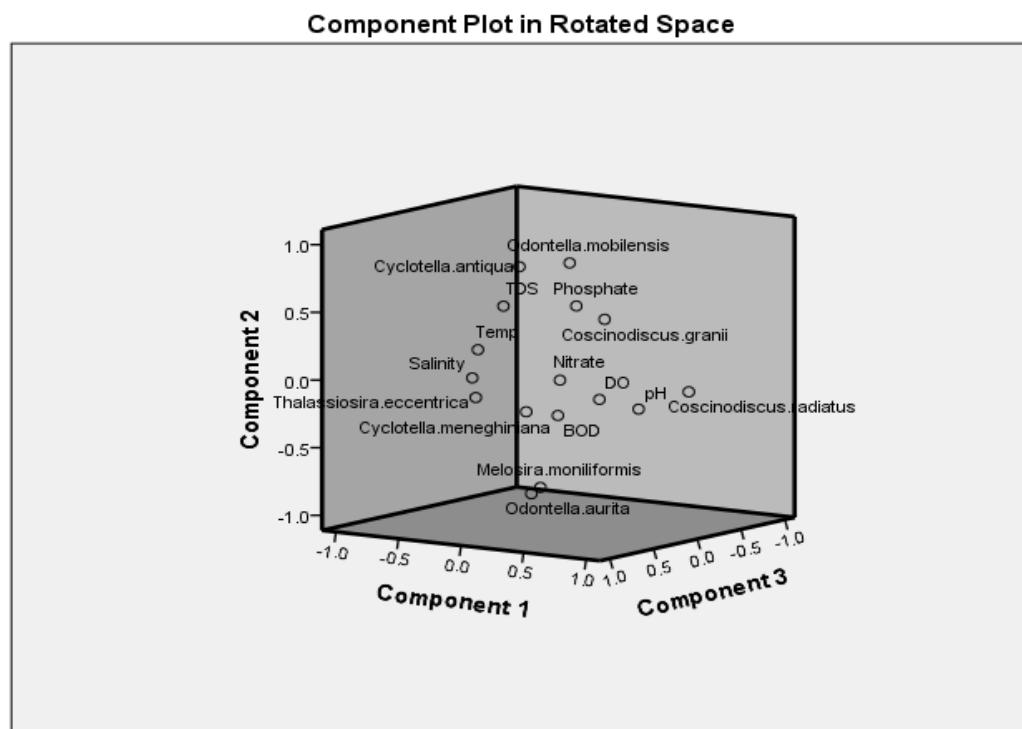


Figure 4 PCA Analysis of harmful centric diatoms with environmental parameters of the upper reaches of the Bonny Estuary

Figure 5 showed the interaction between the environmental factors and harmful pennate diatom species *N. vermicularis* and *Amphiprora* species showed a strong positive correlation with DO and BOD. *B. paxillifera* shows a strong positive correlation with temperature and negative correlation with salinity. *G. accumunatum* showed positive correlation with phosphate and nitrate while *N. stigma* was slightly correlated with Ph.

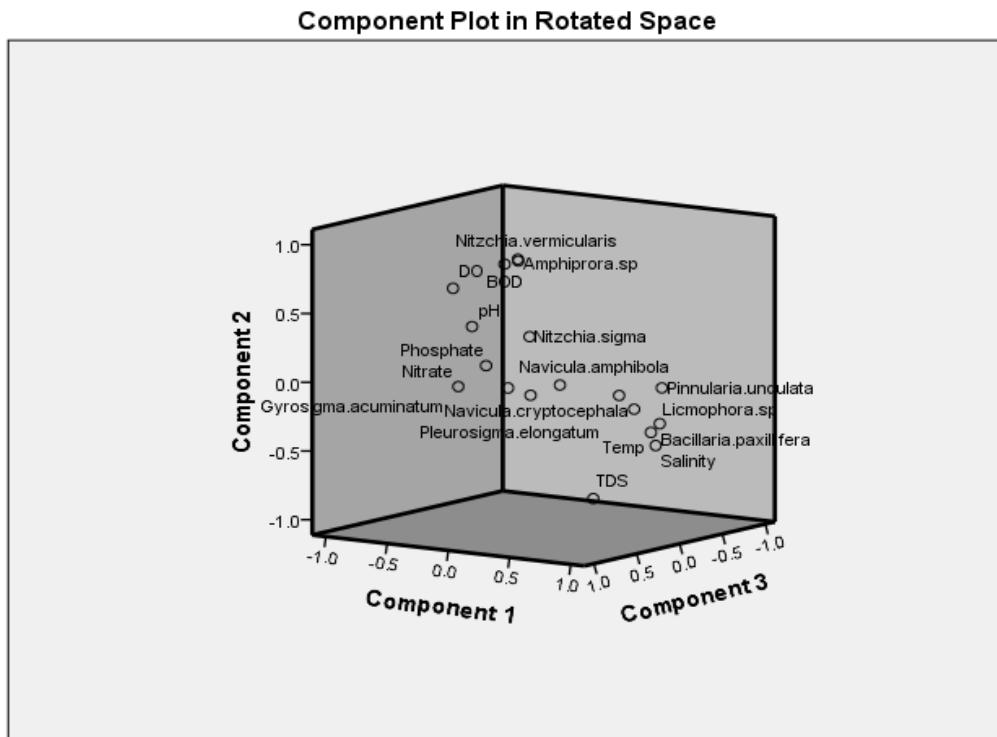


Figure 5 PCA Analysis of pennate harmful diatoms with environmental parameters of the upper reaches of the Bonny Estuary

Table 4 Diversity Indices of Harmful diatoms in the upper reaches of Bonny Estuary

Station	1	2	3
Taxa_S	15	14	15
Individuals	5943	5893	5999
Dominance_D	0.067	0.060	0.068
Shannon_H	2.793	2.818	2.774
Evenness_e^H/S	0.907	0.930	0.890
Margalef	1.903	1.926	1.921

Table 4 shows the diversity indices across the stations. The highest taxa value of 15 was recorded in stations 1 and 3, while the least taxa value of 14 was recorded in station 2. The number of individuals reported the highest value of 5999 in station 3 while the lowest value of 5893 was reported in station 2. Dominance_D with the highest value of 0.068 was recorded in station 3, followed by 0.067 in station 1 while the lowest value of 0.060 was recorded in station 2. The Shannon index and evenness recorded the highest values of 2.818 in station 2, while the lowest values of 2.774 in station 3. The highest Margalef value of 1.926 was recorded in station 2 and the least value of 1.903 was recorded in station 1.

4. DISCUSSION

The pH values recorded in this study area were well within the neutral preferred pH recommended by the World Health Organization, (2008) for optimal fish and aquatic life. The mean temperature values across the Stations were within the observed normal range with reference to the locations in the Niger Delta region. Abowei, (2010) reported a temperature range of between 27°C to 31°C, Omokheyeyeke, (2014) and Jamabo, (2007) also reported a temperature range between 27°C and 30°C in the upper

Bonny River of the Niger Delta. The Dissolved Oxygen (DO) values in the stations were higher at the upper reaches sampled station than the lower reaches but varied slightly in the central reaches, according to Dienye and Sikoki, (2019).

Salinity values recorded in this study were lower than the range reported by Dienye et al., (2022) in the central bonny estuary and Chindah and Nduaguibe, (2003) of $11.5\pm1.8\%$ to $20.3\pm3.0\%$ for the Lower Bonny River. This trend of fluctuation could be attributed to freshwater influx from adjoining river into the estuary and also effluent water discharge from several industrial establishments carrying out bunkering activities and domestic activities, which are prevalent along the upper, reaches area of the Estuary. The recorded biological oxygen demand is lower than what was reported by Vincent-Akpu and Nwachukwu, (2016) in Nembe and Dienye, (2021) in the bonny central estuary.

Total dissolved solids (TDS) values ranged from 18.31 to 23.05 (mgL^{-1}) Vincent-Akpu and Nwachukwu, (2016) reported TDS values of 13.1 (mgL^{-1}) in Nembe and 14.9 (mgL^{-1}) in Bonny. The higher total dissolved organic solid concentration recorded in this study might be attributed to increased surface runoff, overland flow as well as higher discharge of organic wastes into the estuary. Phosphate values recorded in the study area were lower than results from Dienye, (2021) who reported a higher phosphate value in the central Bonny Estuary. Nitrate values were higher than values were within the range reported by Umesi, (1999) in the upper Bonny Estuary.

The harmful diatom algal species composition was dominated with the highest number of species by *Bacillariaceae*. Pennate diatoms recorded 64% with ten species, while centric diatoms recorded 36% with eight species. The result was similar to other research indicating that *Bacillariophyceae* is the dominant genera in the water sample (Bazin et al., 2014). Eight species of centric diatoms were recovered in this study, while Krammer and Lange-Bertalot, (1991) reported seven centric diatoms which belong to species with widespread distribution in the Holarctic.

Onyema et al., (2003) investigated the diatoms phytoplankton in an estuarine creek in Lagos and recorded a total of 18 species of centric diatoms. Dissanayake et al., (2021) recorded eight harmful diatoms from the Southern Coast of Sri Lanka. Harmful diatoms are a common element of epipelagic communities. It is recognized that diatoms diversity is sensitive to environmental variables and responds to changing environmental conditions and that their community structure may quickly respond to changing physical, chemical and biological conditions in the environment (Mooser et al., 1996).

It was recorded in this study that salinity and temperature showed a strong positive correlation with pennate diatoms, while pH, nitrate and phosphate showed a strong correlation with harmful centric diatoms. TDS showed a negative correlation for harmful centric diatoms. In the estuarine system, low cell biomass may be due to the high turbidity levels hence fluctuating photochemistry (Pierce et al., 2004; Shirota, 1989). For the nearshore stations, the low cell biomass of these harmful algae may have resulted from tidal movement as centric diatoms have flagellated gametes but are otherwise swept passively by currents, while pennate diatoms are capable of limited motility. This could be the result of low density of harmful centric diatoms in the upper reaches of the estuary.

The mean density per litre of harmful diatoms revealed that *Thalassiosira eccentrica* species recorded the least density value (318.33CellsL^{-1}) and *Cyclotella meneghiniana* recorded the highest density value ($1146.67\text{CellsL}^{-1}$) for harmful centric diatoms across the sampling stations. *Nitzchia vermicularis* recorded the least density value (602.34CellsL^{-1}) and *Navicula amphibola* recorded the highest density value ($2022.34\text{CellsL}^{-1}$) for pennate harmful diatoms across the sampled stations. This contradicts the findings of Kiteresi et al., (2013) who recorded a low mean density value of harmful centric *Thalassiosira* species in Kenya Coast. *Odontella aurita* recorded as the highest density value across the station, Usup et al., (2002) reported the cell densities of HABs species recorded were not high enough ($<10^6$ cells/L for non-toxic production species and $<10^3$ cells/L for toxic production species) to be considered blooms which are in agreement with the density values of harmful diatoms in the study area.

The composition and diversity of diatom communities are controlled by the biotic and abiotic environment (e.g., the abundance of grazers, nutrients and habitat characteristics), the environmental preferences and tolerances of different diatom species are relatively narrow (Soininen, 2007). Diversity indices are parameters used to indicate the level of stability of an observed community structure, which is closely related to the habitat characteristics occupied by the biota. In this study, the diversity indices of harmful diatoms slightly varied across stations. According to the classification of the Shannon-Wiener index, if the diversity index is lower than 1, then the biota communities would be regarded as unstable.

In contrast a diversity index of 1-3 would be considered moderately unstable and a value higher than three would signify a prime condition (Mokoginta, 2016). It means that the diversity index across the stations was considered relatively stable. Dominance_D index ranges from 0-1, meaning moderately dominance since the value was slightly above 1. Ofonmbuk and Lawrence, (2015) reported a higher Margalef's diversity valve of 2.871 to 3.513 in the Qua-iboe river estuary. The low diversity

values could be attributed to the effects of stress conditions in the environment and also the impact of bunker activities, which are highly probable in the study area, as reported by Adesalu and Nwankwo, (2008).

Diversity is dependent on a vital ecological process such as competition, predation and succession and therefore changes in these processes can alter the species diversity index through changes in evenness (Stirling and Wilsey, 2001). The distribution of harmful diatoms species (Bacillariophyceae) is known to develop harmful algae blooms that increasingly affect aquaculture and tourism. The research on Bacillariophyceae to identify species diversity that caused harmful algae blooming was pursued by several researchers (Ruggiero et al., 2015). Consequently, the cell densities of other harmful diatom species recovered in the upper reaches of the Estuary cannot be treated as blooms but has significant tendencies of possible bloom formation if the environmental factors are not adequately checked.

5. CONCLUSION

The present study revealed the presence of harmful (centric and pennate) diatoms in the upper reaches of the Bonny Estuary study. In this study, the abundance and distribution of these harmful diatoms are a significant issue that affects the health of aquatic ecosystems and poses a threat to human activities such as fishing and recreation. In this study, the least abundant species was *Thalassiosira eccentrica*, while *Cyclotella meneghiniana* recorded the highest abundance for harmful centric diatoms. *Nitzchia vermicularis* recorded the least abundance and *Navicula amphibola* recorded the highest abundance of pennate harmful diatoms across the sampled stations.

Salinity and temperature showed a strong positive correlation with pennate diatoms, while pH, nitrate, and phosphate showed a strong correlation with harmful centric diatoms. The diversity indices showed that all species were evenly spread across all stations and it is therefore recommended further research is necessary to develop effective management strategies that can mitigate the possible formation of blooms and their harmful effects in the upper reaches of the Bonny Estuary.

Informed consent

Not applicable.

Ethical approval

The ethical guidelines are followed in the study for species observation & identification.

Conflicts of interests

The authors declare that there are no conflicts of interests.

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Data and materials availability

All data associated with this study are present in the paper.

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